REMARKS

STATUS OF THE CLAIMS

1. (Original) A method of determining the binding specificity of a ligand to a cell surface moiety, the method comprising the steps of:

(a) providing one or more cell types such that each cell type has a different membrane-anchored electrophoretic probe, each membrane-anchored electrophoretic probe having a membrane anchoring moiety connected by a cleavable linkage to an electrophoretic tag, said tag having distinct optical or electrophoretic properties with respect to electrophoretic tags of other cell types;

(b) combining the one or more cell types with a ligand, and

(c) exposing the cell types and ligand to conditions such that at least one cleavable linkage is cleaved in at least one membrane-anchored electrophoretic probe of cells having a cell surface moiety to which said ligand is bound, whereby one or more electrophoretic tags are released; and

(d) electrophoretically separating and identifying the one or more released electrophoretic tags, to determine the specificity of the ligand for the cell surface moieties of the cell types.

2. (Original) The method of claim 1, wherein said exposing comprises

attaching to each cell having a bound ligand, a proximity-dependent cleavage inducing group, such that each said probe is cleavable only by such a cleavage inducing group on the same cell surface as the probe, and

activating said cleavage inducing group.

3. (Original) The method of claim 2, wherein said probe is cleavable by a short-lived chemical species generated by a sensitizer group, and said exposing includes

attaching such a sensitizer group to each cell having a bound ligand, and

activating the sensitizer group.

4. (Original) The method of claim 3, wherein the sensitizer group is a photosensitizer.

5. (Original) The method of claim 3, wherein the sensitizer group is attached to the ligand.

6. (Original) The method of claim 3, wherein the sensitizer group is attached to a secondary molecule which forms a complex with the ligand.

7. (Original) The method of claim 6, wherein the ligand is an antibody, the sensitizer is conjugated to a secondary antibody immunospecific against the antibody, and said exposing includes adding the secondary antibody and conjugated sensitizer to the cells and bound antibody.

8. (Original) The method of claim 1, wherein said step of combining comprises isolating cell types having at least one ligand bound to at least one cell surface moiety.

9. (Original) The method of claim 8, wherein said cleavable linkage is chemically cleavable, photochemically cleavable, or enzymatically cleavable.

10. (Original) A method of identifying a cell surface antigen specific to substantially only one of a plurality of cell types, the method comprising the steps of:

(a) providing a plurality of cell types with cell surface antigens such that each cell type has a different membrane-anchored electrophoretic probe, the membrane-anchored electrophoretic probe having a membrane anchoring moiety connected by a cleavable linkage to an electrophoretic tag, said tag having distinct optical or electrophoretic properties with respect to electrophoretic tags of other cell types of the plurality;

(b) combining the one or more cell types with a candidate antibody;

(c) exposing the cell types and antibody to conditions such that at least one cleavable linkage is cleaved in at least one membrane-anchored electrophoretic probe of cells having a cell surface antigen to which said antibody is bound, whereby one or more electrophoretic tags are released;

(d) electrophoretically separating and determining the relative quantities of the one or more released electrophoretic tags to determine whether the candidate antibody binds to a cell surface antigen present on substantially only one of the plurality of cell types; and

(e) repeating steps (b)-(d) until a cell surface antigen specific to substantially only one of the plurality of cell types is identified.

11. (Original) The method of claim 10, wherein said exposing comprises

attaching to each cell having a bound antibody, a proximity-dependent cleavage inducing group, such that each said probe is cleavable only by such a cleavage inducing group on the same cell surface as the probe, and

activating said cleavage inducing group.

12. (Original) The method of claim 11, wherein said probe is cleavable by a short-lived chemical species generated by a sensitizer group, and said exposing includes

attaching such a sensitizer group to each cell having a bound antibody, and

activating the sensitizer group.

13. (Original) The method of claim 12, wherein the sensitizer group is a photosensitizer.

14. (Original) The method of claim 13, wherein the sensitizer group is attached to the antibody.

15. (Original) The method of claim 13, wherein the sensitizer is conjugated to a secondary antibody immunospecific against the antibody, and said exposing includes adding the secondary antibody and conjugated sensitizer to the cells and bound antibody.

16. (Original) The method of claim 10, wherein said step of combining comprises isolating cell types having at least one antibody bound to at least one cell surface antigen.

17. (Original) The method of claim 16, wherein said cleavable linkage is chemically cleavable, photochemically cleavable, or enzymatically cleavable.

18. (Original) The method of claim 10, wherein the determining of step (d) comprises measuring the area of tag peaks in an electropherogram of said released tags, and identifying a cell surface antigen specific to substantially only one of the plurality of cell types comprises identifying one test tag peak in said electropherogram that is at least 90% of the sum of the areas of all the test tag peaks in the electropherogram.

19. (Original) The method of claim 18, wherein said one test eTag peak is at least 95% of the sum of the areas of all the test eTag peaks in the electropherogram.

20. (Original) The method of claim 18, wherein identifying a cell surface antigen specific to substantially only one of the plurality of cell types comprises identifying one test tag peak in said electropherogram having an area that is at least twice the area of the next largest test tag peak in the electropherogram.

21. (Original) The method of claim 20, wherein said one test tag peak is at least four times the area of the next largest test tag peak in the electropherogram.

22. (Original) The method of claim 10, further comprising identifying said candidate antibody which binds to said cell surface antigen.

23. (Original) A method of determining the binding affinity of a compound for a cell surface antigen, the method comprising the steps of:

(a) providing one or more test cell-antibody pairs, each such pair comprising (i) a test cell having a membrane-anchored electrophoretic probe, the membrane-anchored electrophoretic probe having a membrane anchoring moiety connected by a cleavable linkage to an electrophoretic tag, said tag having distinct optical or electrophoretic properties with respect to electrophoretic tags of other test cells of the plurality, and (ii) at least one antibody specific for an cell surface antigen of the test cell of such pair, such cell surface antigen being different from cell surface antigens on other test cells of the plurality;

(b) combining the compound with the test cell-antibody pairs under conditions that permit the binding of the antibodies and the compound to one or more cell surface antigens recognized by said compound or antibodies;

(c) exposing the test cell-antibody pairs to conditions such that at least one cleavable linkage is cleaved in at least one membrane-anchored electrophoretic probe of cells having a cell surface antigen to which said antibody is bound, whereby one or more electrophoretic tags are released;

(d) electrophoretically separating the released tags; and

(e) determining the relative quantities of each of the one or more released electrophoretic tags to determine the binding affinity of the compound for the cell surface antigens.

24. (Original) The method of claim 23, wherein said exposing comprises

attaching to each cell having a bound antibody, a proximity-dependent cleavage inducing group, such that each said probe is cleavable only by such a cleavage inducing group on the same cell surface as the probe, and

activating said cleavage inducing group.

25. (Original) The method of claim 24, wherein said probe is cleavable by a short-lived chemical species generated by a sensitizer group, and said exposing includes

attaching such a sensitizer group to each cell having a bound antibody, and

activating the sensitizer group.

26. (Original) The method of claim 25, wherein the sensitizer group is a photosensitizer.

27. (Original) The method of claim 26, wherein the sensitizer group is attached to the antibody.

28. (Original) The method of claim 26, wherein the sensitizer is conjugated to a secondary antibody immunospecific against the antibody, and said exposing includes adding the secondary antibody and conjugated sensitizer to the cells and bound antibody.

29. (Original) The method of claim 23, further comprising the step of comparing the relative quantities obtained in step (e) with those obtained when steps (a), (c) and (d) are carried out in the absence of step (b).

30. (Original) A method of determining the binding specificity of a compound for an internalizing cell surface receptor, the method comprising the steps of:

(a) providing a plurality of test cell-antibody pairs, each such pair comprising (i) a test cell having a membrane-anchored electrophoretic probe, the membrane-anchored electrophoretic probe having a membrane anchoring moiety connected by a cleavable linkage to an electrophoretic tag

having distinct optical or electrophoretic properties with respect to electrophoretic tags of other test cells of the plurality, and (ii) at least one antibody effective to bind to an internalizing cell surface receptor of the test cell of such pair;

(b) combining the compound with the plurality of test cells under conditions that permit the endocytosis of complexes that form between the compound and one or more of the internalizing cell surface receptors;

(c) combining with the test cells the plurality of antibodies under conditions that permit binding to internalizing cell surface receptors;

(d) exposing the test cell-antibody pairs to conditions such that at least one cleavable linkage is cleaved in at least one membrane-anchored electrophoretic probe of cells having an internalizing cell surface receptor to which said antibody is bound, whereby one or more electrophoretic tags are released;

(e) electrophoretically separating the released tags; and

(f) determining the relative quantities of each of the one or more released electrophoretic tags to determine the specificity of the compound for the plurality endocytosing cell surface receptors.

31. (Original) The method of claim 30, wherein said exposing comprises

attaching to each cell having a bound antibody, a proximity-dependent cleavage inducing group, such that each said probe is cleavable only by such a cleavage inducing group on the same cell surface as the probe, and

activating said cleavage inducing group.

32. (Original) The method of claim 30, wherein the internalized cell surface receptor of each test cell is different from internalizing cell surface receptors on other test cells of the plurality.

33. (Original) The method of claim 30, wherein said probe is cleavable by a short-lived chemical species generated by a sensitizer group, and said exposing includes

attaching such a sensitizer group to each cell having a bound antibody, and

activating the sensitizer group.

34. (Original) The method of claim 33, wherein the sensitizer group is a photosensitizer.

35. (Original) The method of claim 33, wherein the sensitizer group is attached to the antibody.

36. (Original) The method of claim 33, wherein the sensitizer is conjugated to a secondary antibody immunospecific against the antibody, and said exposing includes adding the secondary antibody and conjugated sensitizer to the cells and bound antibody.

37. (Original) The method of claim 30, further comprising the step of comparing the relative quantities obtained in step (f) with those obtained when steps (a) and (c)-(e) are carried out in the absence of step (b).